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Comparison of structure and composition of bacterial core communities in mature drinking water biofilms and bulk water of a local network

SUPPLEMENTAL MATERIAL

(revised version November 26, 2011)

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Running title: *Bacteria in mature drinking water biofilms*

1 **Supplementary Table S1.** List of bulk water samples and their main properties. No chlorine residues were detected in all samples.

2 Legend: n.d., not determined.

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Designation	Sampling location	Sampling Date	Sample origin	Water circulation	CFU/ml (R2A, 20°C, 48h)	CFU/ml (R2A, 36°C, 48h)	CFU/ml (R2A, 20°C, 72h)	CFU/ml (R2A, 36°C, 72h)	Bacterial cell counts/ml [10 ⁵ cells]	temperature [°C]	pH	conductivity (µS)
T-HZI-1	HZI M-build.	June 23, 2009	tap	municipal water	2.3	1.0	10.0	10.7	1.25	13.1	8.7	155
T-HZI-2	HZI E-build.	June 23, 2009	receiver tank	municipal water	0.3	5.7	0.3	5.7	2.35	14.5	8.6	157
T-HZI-3	HZI D-build.	June 23, 2009	room D0.04 tap	main circulation	0.3	0.0	0.7	0.0	2.57	12.5	8.5	158
T-HZI-4	HZI Y-build.	June 23, 2009	room Y4.19 tap	main circulation	n.d.	17.0	n.d.	17.0	1.95	20.8	8.2	156
T-HZI-5	HZI E-build.	June 23, 2009	reverse osmosis concentrate	main circulation	8.0	18.7	35.7	24.7	1.42	16.3	8.2	715
T-BS-1	Inner city of Braunschweig	June 24, 2009	tap	municipal water	6.7	30.3	n.d.	n.d.	2.54	23.7	8.3	128
T-BS-2	Inner city of Braunschweig	June 24, 2009	tap	municipal water	2.0	556.7	n.d.	n.d.	2.00	15.6	8.3	164

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1 **Supplementary Table S2.** Overview about the biofilm samples and their main properties.

Designation	Sampling location	Sampling Date	Sample origin	Material	Water circulation	Colour	Age (years)
B-HZI-1	HZI L-build.	May 7, 2009	prefilter	PVC	main circulation	brown	>20
B-HZI-2	HZI D-build.	June 24, 2009	tube at the tap	teflon®	main circulation	yellow beige	>7
B-HZI-3	HZI L-build.	May 14, 2009	main tube	copper	looped fire water mains	green	>20
B-HZI-4	HZI L-build.	May 14, 2009	water meter	stainless steel	looped fire water mains	brown	>20
B-HZI-5	HZI L-build.	May 14, 2009	control window	stainless steel	looped fire water mains	brown	>20
B-HZI-6	HZI L-build.	May 14, 2009	control window	glass	looped fire water mains	brown	>20
B-HZI-7	HZI M-build.	June 23, 2009	receiver tank	stainless steel	municipal water	brown	unknown
B-BS-1	Inner city of Braunschweig	June 24, 2009	flush water container	PVC	municipal water	brown	3

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1 **Supplementary Table S3.** Taxonomic identification of phylotypes (PT) retrieved from bulk water (bands excised from SSCP fingerprints shown in
 2 Fig. 3a.) Legend: Sample origin: D, PT only observed in DNA-based fingerprints; R, only observed in RNA-based fingerprints; DR, observed in
 3 DNA and RNA-based fingerprints; ^{HWW}, Phylotype already observed in previous studies on HWW drinking water system (distinction between
 4 sequences: *, distinct less or equal to 3 nt; **, less or equal to 6nt; distinction after correction for errors) (Eichler et al. 2006, Kahlisch et al. 2011); ^a,
 5 only tentative phylogenetic assignment due to reduced sequence information (short or sequence with many ambiguous bases, or no related sequence
 6 in data bases).

Phylotype designation	Sample origin	GenBank accession no.	Taxonomic group	Closest 16S rRNA gene sequence (Accession no.)	source of closest sequence	% Similarity	Closest described species (Accession no.)	% Similarity
T015 ^a	D	FR796669	Actinobacteria	Uncultured actinobacterium clone CB31D05 16S ribosomal RNA gene, partial sequence (EF471701)	whole surface water from Chesapeake bay	89%	Tetrasphaera veronensis (Y14596)	87%
T016 ^{HWW*}	D	FR796670	Actinobacteria	Uncultured bacterium clone GC1m-4-84 16S ribosomal RNA gene, partial sequence (EU640899)	Lake Michigan	94%	Brevibacterium albus (EF158852)	86%
T017 ^a	D	FR796671	Actinobacteria	Uncultured bacterium isolate SSCP band GR1-D2-1_8 16S ribosomal RNA gene, partial sequence (DQ077614)	drinking water distribution system HWW	88%	Candidatus Planktophila limnetica (FJ428831)	91%
T018	D	FR796672	Actinobacteria	Uncultured actinobacterium clone I-DW-40 16S ribosomal RNA gene, partial sequence (GQ453106)	drinking water	94%	Candidatus Planktophila limnetica (FJ428831)	93%
T019	D	FR796673	Actinobacteria	Uncultured Actinomycetales bacterium clone Gap-2-37 16S ribosomal RNA gene, partial sequence (EU642138)	Milwaukee harbor	95%	Candidatus Planktophila limnetica (FJ428831)	93%
T020 ^{HWW*}	D	FR796674	Actinobacteria	Uncultured actinobacterium clone TR1F1 16S ribosomal RNA gene, partial sequence (EU117957)	lake epilimnion	98%	Candidatus Planktophila limnetica (FJ428831)	96%
T021 ^{HWW**}	D	FR796675	Actinobacteria	Uncultured bacterium clone CABC1F12 16S ribosomal RNA gene, partial sequence (GU127254)	aphotic layer; anoxic zone	94%	Candidatus Planktophila limnetica (FJ428831)	91%

T003 ^{HWW*}	D	FR796657	Alphaproteobacteria	Uncultured bacterium isolate SSCP band DNA1-12-14 16S ribosomal RNA gene, partial sequence (DQ077623)	drinking water distribution system HWW*	100%	Candidatus Pelagibacter ubique (EU410957)	86%
T004 ^{HWW*}	D	FR796658	Alphaproteobacteria	Uncultured bacterium isolate SSCP band DNA1-12-14 16S ribosomal RNA gene, partial sequence (DQ077624)	drinking water distribution system HWW	98%	Ehrlichia ewingii (U96436)	84%
T026	R	FR796680	Alphaproteobacteria	Uncultured alpha proteobacterium clone AKYH1214 16S ribosomal RNA gene, partial sequence (AY921890)	farm soil adjacent to a silage storage	90%	Prosthecomicrobium consociatum (FJ560750)	90%
T027	R	FR796681	Alphaproteobacteria	Bosea sp. 7 GUW 16S ribosomal RNA gene, partial sequence (EU496542)	water; natural oil seeps	99%	Bosea eneeae (EF519707)	99%
T028 ^a	R	FR796682	Alphaproteobacteria	Bosea sp. RA62 16S ribosomal RNA gene, partial sequence (FJ898313)	spring	89%	Bosea thiooxidans (EU730912)	88%
T002 ^a	D	FR796656	Bacteroidetes	Uncultured bacterium clone G910P35FB10.T0 16S ribosomal RNA gene, partial sequence (EU172242)	air	84%	Sediminibacterium ginsengisoli (EF067860)	84%
T035	D	FR796689	Bacteroidetes	Uncultured bacterium clone TLM10/TLMdgg01 16S ribosomal RNA gene, partial sequence (AF534434)	Toolik Lake main station at 3 m depth	97%	Adhaeribacter aquaticus (AJ626894)	85%
T036 ^{HWW*}	D	FR796690	Bacteroidetes	Uncultured bacterium isolate SSCP band GR1-RNA1-3-27 16S ribosomal RNA, partial sequence (DQ077593)	drinking water distribution system HWW	99%	Sejongia jeonii (AY553294)	83%
T037	R	FR796691	Bacteroidetes	Uncultured Bacteroidetes bacterium partial 16S rRNA gene, clone JG35-K2-AG43 (AM403313)	soil	93%	Flexibacter canadensis (AB078046)	85%
T038	D	FR796692	Bacteroidetes	Uncultured Bacteroidetes bacterium clone IRD18D04 16S ribosomal RNA gene, partial sequence (AY947930)	USA: Massachusetts, Ipswich River	95%	Ekhidna lutea (AM746475)	86%
T039 ^a	D	FR796693	Bacteroidetes	Uncultured bacterium isolate SSCP band TW16-D_14_9 16S ribosomal RNA gene, partial sequence (DQ077625)	drinking water distribution system HWW	88%	Sediminibacterium salmoneum (EF407879)	87%
T040	D	FR796694	Bacteroidetes	Uncultured Bacteroidetes bacterium partial 16S rRNA gene, clone NE02 (AJ575726)	Grosse Fuchskuhle	90%	Sediminibacterium ginsengisoli (EF067860)	90%
T041	D	FR796695	Bacteroidetes	Uncultured Sphingobacteria bacterium clone LW9m-1-10 16S ribosomal RNA gene, partial sequence (EU641456)	Lake Michigan	98%	Terrimonas ferruginea (AM230484)	93%

T042 ^{HWW**}	D	FR796696	Bacterioidetes	Uncultured bacterium isolate SSCP band TW16-D_14_9 16S ribosomal RNA gene, partial sequence (DQ077625)	drinking water distribution system HWW	93%	Sediminibacterium salmoneum (EF407879)	92%
T043 ^{HWW*}	D	FR796697	Bacterioidetes	Uncultured bacterium isolate SSCP band TW16-D_14_9 16S ribosomal RNA gene, partial sequence (DQ077625)	drinking water distribution system HWW	100%	Sediminibacterium salmoneum (EF407879)	97%
T044 ^{HWW*}	D	FR796698	Bacterioidetes	Uncultured bacterium isolate SSCP band TW16-D_14_9 16S ribosomal RNA gene, partial sequence (DQ077625)	drinking water distribution system HWW	98%	Sediminibacterium salmoneum (EF407879)	96%
T001 ^{HWW*}	R	FR796655	Betaproteobacteria	Uncultured bacterium isolate SSCP band GT-8R_03-20 16S ribosomal RNA, partial sequence (DQ077557)	drinking water distribution system HWW	97%	Candidatus Tremblaya princeps (AF476079)	97%
T007 ^a	D	FR796661	Betaproteobacteria	Uncultured beta proteobacterium partial 16S rRNA gene, isolate DGGE band WETLE-13B (FM991990)	constructed wetland	87%	Candidatus Accumulibacter phosphatis (AY962316)	87%
T008 ^{HWW*}	D	FR796662	Betaproteobacteria	Uncultured bacterium clone DP10.5.4 16S ribosomal RNA gene, partial sequence (FJ612426)	lake water	100%	Methylophilus methylotrophus (GQ175365)	96%
T009 ^{HWW*}	R	FR796663	Betaproteobacteria	Nitrosospira briensis 16S ribosomal RNA gene, partial sequence (AY123800)	rhizosphere	100%	Nitrosospira briensis (AY123800)	100%
T010	R	FR796664	Betaproteobacteria	Uncultured bacterium partial 16S rRNA gene, clone SZB2 (AM176880)	mangrove sediment	98%	Oxalicibacterium flavum (AY061962)	94%
T011 ^{HWW*}	D	FR796665	Betaproteobacteria	Uncultured beta proteobacterium clone PRD18F04 16S ribosomal RNA gene, partial sequence (AY948047)	USA: Massachusetts, Parker River	99%	Acidovorax facilis (EU730927)	99%
T012 ^{HWW*}	R	FR796666	Betaproteobacteria	Uncultured bacterium isolate SSCP band RNA2-9-10 16S ribosomal RNA, partial sequence (DQ077559)	drinking water distribution system HWW	96%	Acidovorax facilis (GQ284412)	95%
T013	D	FR796667	Betaproteobacteria	Uncultured beta proteobacterium clone CB31D01 16S ribosomal RNA gene, partial sequence (EF471693)	whole surface water from Chesapeake bay	97%	Kerstersonia gyiorum (AY131213)	97%
T014 ^a	D	FR796668	Betaproteobacteria	Uncultured Polynucleobacter sp. isolate SSCP band 155-0-5 16S ribosomal RNA gene, partial sequence (GU088519)	water sample from Sumauma river	86%	Polynucleobacter necessarius subsp. asymbioticus (CP001010)	86%
T029 ^{HWW*}	R	FR796683	Cyanobacteria	Uncultured bacterium isolate SSCP band TW15-RNA1-14-2 16S ribosomal RNA, partial sequence (DQ077556)	drinking water distribution system HWW	100%	Gracilibacter thermotolerans (DQ117469)	85%

T030 ^{HWW*}	R	FR796684	Cyanobacteria	Uncultured bacterium isolate SSCP band PT_27 16S ribosomal RNA gene, partial sequence (GQ917147)	drinking water distribution system HWW	93%	Megamonas rupellensis (EU346729)	81%
T031 ^a	R	FR796685	Cyanobacteria	Uncultured bacterium isolate SSCP band PT_27 16S ribosomal RNA gene, partial sequence (GQ917147)	drinking water distribution system HWW	87%	Methylobacter tundripaludum (AJ414655)	85%
T032	R	FR796686	Gammaproteobacteria	Methylobacter tundripaludum 16S ribosomal RNA, type strain SV96T (AJ414655)	iron-rich snow	99%	Methylobacter tundripaludum (AJ414655)	99%
T033 ^{HWW*}	R	FR796687	Gammaproteobacteria	Uncultured bacterium isolate SSCP band RNA2-8-7 16S ribosomal RNA, partial sequence (DQ077602)	drinking water distribution system HWW	99%	Methylocaldum gracile (U89298)	92%
T022	R	FR796676	Nitrospira	Uncultured Nitrospira sp. clone I-GAC-2 16S ribosomal RNA gene, partial sequence (GQ452974)	drinking water	93%	Nitrospira moscoviensis (X82558)	91%
T005	D	FR796659	Planctomycetes	Uncultured bacterium gene for 16S rRNA, partial sequence, clone: MIZ10 (AB179501)		89%	Mycoplasma dispar (AF412979)	83%
T006 ^a	D	FR796660	Planctomycetes	Uncultured bacterium isolate SSCP band GT-8R_03-21 16S ribosomal RNA, partial sequence (DQ077597)	drinking water distribution system HWW	85%	Nevskia ramosa (AJ001011)	88%
T034	R	FR796688	Planctomycetes	Uncultured bacterium clone N1-103 16S ribosomal RNA gene, partial sequence (EU443041)	Nam Co Lake water	99%	Gemmata obscuriglobus (X85248)	87%
T023 ^a	R	FR796677	TM6	Uncultured bacterium gene for 16S ribosomal RNA, partial sequence, isolate: DGGE band: 8 (AB472269)	crude oil-contaminated soil	82%	Candidatus Glomeribacter gigasporarum (AM889131)	87%
T024 ^{HWW**}	R	FR796678	TM6	Uncultured bacterium isolate SSCP band PT_19 16S ribosomal RNA gene, partial sequence (GQ917139)	drinking water distribution system HWW	94%	Desulfococcus biacutus (AJ277887)	88%
T025	R	FR796679	TM6	Uncultured bacterium clone SGSH795 16S ribosomal RNA gene, partial sequence (GQ347607)	Saanich Inlet, 215 m depth	93%	Nevskia ramosa (AJ001343)	87%

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2 **Supplementary Table S4.** Taxonomic identification of phylotypes from biofilms (excised from SSCP fingerprints shown in Fig. 4a.). Legend as in
3 Table S3.

Phylotype designation	Sample origin	GenBank accession no.	Taxonomic group	Closest 16S rRNA gene sequence (Accession no.)	source of closest sequence	% Similarity	Closest described species (Accession no.)	% Similarity
B016	R	FR796558	Acidobacteria	Uncultured bacterium gene for 16S rRNA, partial sequence, clone: 1333 (AB286500)	activated sludge	99%	Candidatus Solibacter usitatus Ellin6076 (CP000473)	93%
B017	R	FR796559	Acidobacteria	Uncultured bacterium 16S rRNA gene, clone D14305 (AJ617855)	oxic-anoxic interphase of flooded paddy soil	99%	Candidatus Solibacter usitatus (GQ287529)	91%
B048 ^a	D	FR796590	Acidobacteria	Uncultured bacterium clone FFCH3013 16S ribosomal RNA gene, partial sequence (EU132279)	soil from an undisturbed mixed grass prairie reserve	86%	Eubacterium siraeum (EU266550)	87%
B049	R	FR796591	Acidobacteria	Uncultured bacterium clone FFCH3013 16S ribosomal RNA gene, partial sequence (EU132280)	soil from an undisturbed mixed grass prairie reserve	91%	Tepidanaerobacter syntrophicus (AB106354)	88%
B050 ^a	R	FR796592	Acidobacteria	Uncultured bacterium clone 4 16S ribosomal RNA gene, partial sequence (EU362133)	dune sand	88%	Candidatus Desulforudis audaxviator MP104C (CP000860)	85%
B052 ^a	R	FR796594	Acidobacteria	Uncultured bacterium clone AK1DE1_02D 16S ribosomal RNA gene, partial sequence (GQ396963)	soil	77%	Agrobacterium tumefaciens (AF508099)	76%
B054	D	FR796596	Acidobacteria	Uncultured bacterium clone AK1AB1_12H 16S ribosomal RNA gene, partial sequence (GQ396847)	soil	97%	Selenomonas sputigena (GQ422723)	91%
B057	D	FR796599	Acidobacteria	Uncultured bacterium clone 099.F24 16S ribosomal RNA gene, partial sequence (EU355855)	agricultural soil treatment	92%	Desulfovibrio giganteus (AF418170)	88%
B058 ^a	D	FR796600	Acidobacteria	Uncultured delta proteobacterium clone MPWIC_G06 16S ribosomal RNA gene, partial sequence (EF414165)	sponge	89%	Desulfovibrio indonesiensis (Y09504)	85%
B033	D	FR796575	Actinobacteria	Uncultured bacterium clone LL141-8C18 16S ribosomal RNA gene, partial sequence (FJ675406)	feedlot surface material	98%	Thermoleophilum album (AJ458462)	88%
B083	D	FR796625	Actinobacteria	Uncultured bacterium clone TX5A_90 16S ribosomal RNA gene (FJ152798)	alkaline saline soils of the former lake Texcoco	100%	lamia majanohamensis (AB360448)	92%

B084	D	FR796626	Actinobacteria	Uncultured bacterium clone TX5A_90 16S ribosomal RNA gene (FJ152798)	alkaline saline soils of the former lake Texcoco	90%	<i>lamia majanohamensis</i> (AB360448)	83%
B085	D	FR796627	Actinobacteria	Uncultured bacterium clone LL141-8C18 16S ribosomal RNA gene (FJ675406)	feedlot surface material	98%	<i>Thermoleophilum album</i> (AJ458462)	88%
B010 ^a	R	FR796552	Alphaproteobacteria	Uncultured alpha proteobacterium clone GASP-MA4W2_F12 16S ribosomal RNA gene, partial sequence (EF664356)	grassland	78%	<i>Skermanella aerolata</i> strain 5416T-32 (DQ672568)	72%
B011	R	FR796553	Alphaproteobacteria	Uncultured soil bacterium clone Bact.dry.ACETF05 16S ribosomal RNA gene, partial sequence (GU375766)	oil-field soil	94%	<i>Candidatus Reyranela massiliensis</i> (EF394922)	89%
B013 ^a	D	FR796555	Alphaproteobacteria	<i>Wolbachia</i> endosymbiont of <i>Cubitermes</i> sp. clone T5 16S ribosomal RNA gene, partial sequence (EF417899)	<i>Cubitermes</i> sp	78%	<i>Wolbachia pipientis</i> strain wHa (DQ235279)	77%
B021	R	FR796563	Alphaproteobacteria	Uncultured bacterium clone F126 16S ribosomal RNA gene, partial sequence (FJ348594)	activated sludge	98%	<i>Clostridium sporogenes</i> (L09175)	91%
B059 ^a	D	FR796601	Alphaproteobacteria	Uncultured Alphaproteobacteria bacterium 16S rRNA gene from clone QEDQ2AE01 (CU923390)	municipal wastewater sludge	64%	<i>Serratia marcescens</i> (FM213393)	88%
B061	D	FR796603	Alphaproteobacteria	Uncultured bacterium gene for 16S rRNA, partial sequence, clone:14s (AB154445)	PCR-derived sequence from meromictic lake sediment	93%	<i>Desulfomonile limimaris</i> (AF282177)	91%
B062 ^a	D	FR796604	Alphaproteobacteria	Uncultured alpha proteobacterium partial 16S ribosomal RNA, clone SSCP ribotype S-RS-13b (AM989610)	soddy-podzolic soil casts of earthworms	89%	<i>Mesorhizobium thiogangeticum</i> (AJ864462)	86%
B063 ^a	D	FR796605	Alphaproteobacteria	Uncultured alpha proteobacterium partial 16S ribosomal RNA, clone SSCP ribotype S-RS-13b (AM989610)	soddy-podzolic soil casts of earthworms	87%	<i>Pseudoxanthobacter soli</i> (EF465533)	85%
B064 ^a	D	FR796606	Alphaproteobacteria	<i>Hyphomicrobium</i> sp. D3 16S ribosomal RNA gene, partial sequence (EF079086)	ditch sediment collected in freshwater	88%	<i>Azospirillum brasilense</i> (Z29617)	90%
B065	R	FR796607	Alphaproteobacteria	Bacterium Fuku2-ISO-153 16S ribosomal RNA gene, partial sequence (EU409478)	Lake Fuchskuhle	92%	<i>Hyphomicrobium facile</i> (Y14312)	88%
B066	RD	FR796608	Alphaproteobacteria	Uncultured bacterium clone 1C227168 16S ribosomal RNA gene, partial sequence (EU799556)	Newport Harbour, RI	99%	<i>Rhodobacter blasticus</i> (DQ342322)	97%

B067	R	FR796609	Alphaproteobacteria	Uncultured bacterium clone 1C227168 16S ribosomal RNA gene, partial sequence (EU799556)	Newport Harbour, RI	99%	Rhodobacter capsulatus (DQ342320)	98%
B068 ^a	R	FR796610	Alphaproteobacteria	Uncultured bacterium clone nbw1075f08c1 16S ribosomal RNA gene, partial sequence (GQ052877)	skin, volar forearm	85%	Rhodobacter blasticus (DQ342322)	84%
B069	R	FR796611	Alphaproteobacteria	Uncultured bacterium clone H07_SE4A 16S ribosomal RNA gene, partial sequence (FJ592547)	wetland soil	93%	Rhodovulum marinum (AM696693)	91%
B070 ^a	R	FR796612	Alphaproteobacteria	Uncultured alpha proteobacterium partial 16S rRNA gene, clone h5-3 (FN594682)	biofilm from gold mine in Zloty Stok	89%	Woodsholea maritima (AJ578477)	86%
B071	RD	FR796613	Alphaproteobacteria	Uncultured bacterium gene for 16S rRNA, partial sequence, clone: SWB05 (AB294316)	stream	100%	Novosphingobium stygium (U20775)	94%
B072 ^a	D	FR796614	Alphaproteobacteria	Uncultured bacterium partial 16S rRNA gene, isolate DGGE_band_ag8 (AM749502)	lysimeter soil	88%	Anderseniella baltica (AM712634)	89%
B073	D	FR796615	Alphaproteobacteria	Uncultured alpha proteobacterium partial 16S rRNA gene, clone d5-3 (FN594655)	biofilm from gold mine in Zloty Stok	97%	Pedomicrobium australicum (FM886896)	94%
B074	RD	FR796616	Alphaproteobacteria	Uncultured bacterium clone WW1_b1 16S ribosomal RNA gene, partial sequence (GQ264217)	simulated low level waste site	94%	Hyphomicrobium sulfonivorans (AY468372)	97%
B075	D	FR796617	Alphaproteobacteria	Uncultured bacterium clone WW1_b1 16S ribosomal RNA gene (GQ264217)	simulated low level waste site	94%	Hyphomicrobium sulfonivorans (AY468372)	93%
B076	R	FR796618	Alphaproteobacteria	Uncultured alpha proteobacterium partial 16S rRNA gene, clone e10-1 (FN594694)	biofilm from gold mine in Zloty Stok	92%	Hyphomicrobium zavarzinii (Y14306)	91%
B077	R	FR796619	Alphaproteobacteria	Uncultured alpha proteobacterium clone as2-53 16S ribosomal RNA (GU257609)	activated sludge in a membrane bioreactor	92%	Hyphomicrobium sulfonivorans (AY468372)	90%
B078	D	FR796620	Alphaproteobacteria	Uncultured bacterium clone Bul1ah07 16S ribosomal RNA gene (FJ228809)	host:Bulinus africanus	96%	Bradyrhizobium yuanmingense (FJ785218)	92%
B079	D	FR796621	Alphaproteobacteria	Uncultured alpha proteobacterium clone A23YM01RM small subunit (FJ569518)	soil early snow melt site B, Alpes	92%	Rhodoplanes piscinae (AM712913)	90%
B080	D	FR796622	Alphaproteobacteria	Uncultured bacterium clone TY-R-II-OTU6 16S ribosomal RNA gene (FJ178175)	soil derived from quaternary red clay	93%	Rhodoplanes piscinae (AM712913)	91%

B081 ^a	R	FR796623	Alphaproteobacteria	Uncultured bacterium clone MACA-RR12 16S ribosomal RNA gene (GQ500747)	base level cave stream, Roaring River (sediment)	84%	Rhodobium orientis (D30792)	83%
B082	RD	FR796624	Alphaproteobacteria	Uncultured bacterium gene for 16S rRNA, partial sequence, clone: 136 (AB286364)	activated sludge	98%	Hyphomicrobium sulfonivorans (AY305006)	88%
B109	R	FR796651	Alphaproteobacteria	Uncultured alpha proteobacterium clone J-DW-21 16S ribosomal RNA gene, partial sequence (GQ453317)	Surface Water Treatment Plant	96%	Parvularcula lutaonensis (EU346850)	90%
B089	D	FR796631	Bacteroidetes	Uncultured Flexibacteraceae bacterium clone BL017B17 16S ribosomal (DQ188271)	bronchoalveolar lavage fluid from children with cystic fibrosis	93%	Pontibacter korlensis (GQ503321)	86%
B090	RD	FR796632	Bacteroidetes	Uncultured Bacteroidetes bacterium clone I-GAC-12 16S ribosomal RNA (GQ452967)	drinking water treatment plant	99%	Terrimonas ferruginea (AM230484)	98%
B091	RD	FR796633	Bacteroidetes	Uncultured Bacteroidetes bacterium clone I-GAC-13 16S ribosomal RNA (GQ452968)	drinking water treatment plant	99%	Terrimonas ferruginea (AM230484)	93%
B098	D	FR796640	Betaproteobacteria	Uncultured bacterium clone GB062005_2-35 16S ribosomal RNA gene (GQ487819)	groundwater surface water interface sediments	93%	Azospira restricta (DQ974114)	90%
B099 ^a	D	FR796641	Betaproteobacteria	Sterolibacterium sp. TKU1 partial 16S rRNA gene, strain TKU1 (AM990454)	ultra pure water from an industrial cooling water system	89%	Sideroxydans lithotrophicus (DQ386859)	85%
B100	R	FR796642	Betaproteobacteria	Uncultured Burkholderiales bacterium clone Gap-2-58 16S ribosomal RN (EU642196)	Lake Michigan	100%	Methylibium aquaticum (DQ664244)	99%
B101	RD	FR796643	Betaproteobacteria	Uncultured Burkholderiales bacterium partial 16S rRNA gene, clone B6_93 (AM940846)	glacier moraine	100%	Methylibium fulvum (AB245356)	100%
B102	D	FR796644	Betaproteobacteria	Uncultured bacterium clone AK4DE1_01F 16S ribosomal RNA gene (GQ397030)	soil	98%	Thiobacillus thiophilus (EU685841)	92%

B103	R	FR796645	Betaproteobacteria	Uncultured beta proteobacterium clone D10_37 small subunit ribosomal (EU266802)	tar-oil contaminated aquifer sediments	97%	Burkholderia andropogonis (DQ786950)	95%
B104	RD	FR796646	Betaproteobacteria	Uncultured bacterium clone MYS8 16S ribosomal RNA gene (GU305733)	MiYu reservoir water (oligotrophic lake)	98%	Ultramicrobacter hongkongensis (DQ532120)	97%
B105	D	FR796647	Betaproteobacteria	Uncultured Antarctic bacterium LB3-81 16S ribosomal RNA gene, partial sequence (AF173823)	Permanent Antarctic Lake Ice	97%	Derxia gummosa (AB089482)	94%
B106	R	FR796648	Betaproteobacteria	Uncultured bacterium clone nbw877d04c1 16S ribosomal RNA gene, partial sequence (GQ030383)	skin, elbow	93%	Ralstonia syzygii (AY464966)	93%
B107 ^a	R	FR796649	Betaproteobacteria	Uncultured beta proteobacterium clone GASP-WB2S1_E03 16S ribosomal RNA gene, partial sequence (EF073875)	pasture	78%	Nitrosospira multiformis ATCC 25196 (CP000103)	75%
B001	R	FR796543	Chlamydiales	Uncultured Chlamydiales bacterium clone P-7 16S ribosomal RNA gene, partial sequence (AF364569)	environmental sample	100%	Neochlamydia hartmannellae strain A1Hsp (NR_025037)	90%
B002	R	FR796544	Chlamydiales	Uncultured soil bacterium clone 530-2 small subunit ribosomal RNA gene, partial sequence (AY326519)	soil	93%	Parachlamydia acanthamoebae strain CRIB43 (FJ532291)	91%
B003	R	FR796545	Chlamydiales	Uncultured bacterium clone MABRDTU43 16S ribosomal RNA gene, partial sequence (FJ529996)	autotrophic nitrifying biofilm reactor	99%	Parachlamydia acanthamoebae strain CRIB44 (FJ532291.2)	91%
B004	R	FR796546	Chlamydiales	Parachlamydiaceae bacterium CHSL 16S ribosomal RNA gene, partial sequence (GQ221847)	hartmannellid amoeba SL-2	93%	Parachlamydia acanthamoebae strain CRIB45 (FJ532291.3)	92%
B005	R	FR796547	Chlamydiales	Candidatus Protochlamydia sp. CRIB40 16S ribosomal RNA gene, partial sequence (FJ532293)	biofilm from clarifier	98%	Protochlamydia naegleriophila strain CRIB42 (FJ532295)	96%
B006	R	FR796548	Chlamydiales	Uncultured bacterium gene for 16S rRNA, partial sequence, clone: NG_inoculum_16 (AB518086)	activated sludge	93%	Neochlamydia hartmannellae strain A1Hsp (NR_025037)	92%
B007 ^a	D	FR796549	Chlamydiales	Uncultured Chlamydiae bacterium clone DSM2W1u70 16S ribosomal RNA gene, partial sequence (EU635381)	showerhead swab	84%	Candidatus Fritschea bemisiae strain Falk (AY140910)	81%
B008	RD	FR796550	Chlamydiales	Endosymbiont of Acanthamoeba sp. R18 gene for 16S rRNA, partial sequence (AB506679)	Acanthamoeba sp. R18	90%	Protochlamydia naegleriophila strain CRIB41 (FJ532294)	90%

B009 ^a	D	FR796551	Chlamydiales	Candidatus Protochlamydia sp. cvE12 16S ribosomal RNA gene, partial sequence (FJ976092)	fountain	86%	Parachlamydia acanthamoebae strain CRIB43 (FJ532291)	88%
B020 ^a	R	FR796562	Chlamydiales	Uncultured bacterium gene for 16S rRNA, partial sequence, clone: PltcGammaproteobacterium88 (AB424911)	hydrothermal sulfide structure	88%	Bacillus megaterium (EU910239)	86%
B024	D	FR796566	Chloroflexi	Uncultured Chloroflexi bacterium clone AKYG631 16S ribosomal RNA gene, partial sequence (AY921657)	farm soil adjacent to a silage storage bunker	93%	Thermanaeromonas toyohensis (AB062280)	87%
B025	D	FR796567	Chloroflexi	Uncultured bacterium clone 344.F22 16S ribosomal RNA gene, partial sequence (EU353968)	agricultural soil treatment	100%	Thermaerobacter composti (AB454087)	86%
B026	D	FR796568	Chloroflexi	Uncultured bacterium clone 344.F22 16S ribosomal RNA gene, partial sequence (EU353968)	agricultural soil treatment	91%	Ureibacillus thermophilus (DQ348072)	86%
B108	R	FR796650	Chloroflexi	Uncultured Chloroflexi bacterium partial 16S rRNA gene, clone AMJA2 (AM934855)	hydrocarbon-contaminated soil	91%	Moorella perchloratireducens (EF060194)	91%
B056 ^a	D	FR796598	Deltaproteobacteria	Uncultured bacterium clone C9 G3 16S ribosomal RNA gene, partial sequence (GU366869)	temperate forest soil	87%	Geobacter sulfurreducens (U13928)	84%
B060 ^a	R	FR796602	Deltaproteobacteria	Uncultured delta proteobacterium clone TDNP_USbc97_138_1_18 16S ribosomal RNA gene, partial sequence (FJ516890)	upper sediment	88%	Rhodospirillum sulfurexigens (AM710622)	88%
B012	R	FR796554	Firmicutes	Uncultured bacterium isolate SSCP band PT_27 16S ribosomal RNA gene, partial sequence (GQ917147)	HWW drinking water	95%	Leptolyngbya frigida (AY493574)	87%
B022 ^a	D	FR796564	Firmicutes	Uncultured bacterium clone 1-gw2-su4-12 16S ribosomal RNA gene, partial sequence (DQ981803)	river water	79%	Bacillus licheniformis (X68416)	84%
B023	D	FR796565	Firmicutes	Bacillus sp. PLC9 16S ribosomal RNA gene, partial sequence (FJ973430)	treatment water	91%	Bacillus pichinoty (EU373388)	90%
B051 ^a	R	FR796593	Firmicutes	Uncultured Clostridiaceae bacterium gene for 16S rRNA, clone: dgD-50 (AB264067)	PCR-derived from Dugong feces	89%	Anaerospobacter mobilis (AY534872)	88%
B053 ^a	D	FR796595	Firmicutes	Uncultured bacterium clone 16_14D09 16S ribosomal RNA gene, partial sequence (GQ360313)	left upper lung lobe	87%	Ethanoligenens harbinense (EU639425)	86%

B014	RD	FR796556	Gammaproteobacteria	Uncultured bacterium clone WC3_79 16S ribosomal RNA gene, partial sequence (GQ264139)	simulated low level waste site	91%	<i>Legionella rubrilucens</i> (Z32643)	90%
B015	D	FR796557	Gammaproteobacteria	Uncultured Legionellaceae bacterium isolate DGGE gel band M4-3(I) 16S ribosomal RNA gene, partial sequence (FJ467409)	bovine mastitis milk	95%	<i>Legionella lytica</i> (Z49741)	94%
B034 ^a	D	FR796576	Gammaproteobacteria	Uncultured bacterium clone A1-07 16S ribosomal RNA gene, partial sequence (EU857839)	Ross Sea sediment	82%	<i>Propionivibrio limicola</i> (AJ307983)	83%
B035 ^a	R	FR796577	Gammaproteobacteria	Uncultured gamma proteobacterium clone NE41C01cA 16S ribosomal RNA gene, partial sequence (DQ424446)	microbial mat	85%	<i>Sedimenticola selenatireducens</i> (AF432145)	84%
B055 ^a	D	FR796597	Gammaproteobacteria	Uncultured gamma proteobacterium clone NE36D07cA 16S ribosomal RNA gene, partial sequence (DQ424244)	microbial mat	85%	<i>Hyphomicrobium zavarzinii</i> (Y14306)	85%
B086 ^a	D	FR796628	Gammaproteobacteria	Uncultured bacterium clone C2 A14 16S ribosomal RNA gene (GU366816)	temperate forest soil	88%	<i>Thioalkalivibrio denitrificans</i> (AF126545)	89%
B092 ^a	D	FR796634	Gammaproteobacteria	Uncultured bacterium clone D44 16S ribosomal RNA gene (EU234314)	upstream of Wang Yang River	83%	<i>Panacagrmonas perspica</i> (AB257720)	79%
B093	D	FR796635	Gammaproteobacteria	Uncultured bacterium clone D44 16S ribosomal RNA gene (EU234314)	upstream of Wang Yang River	91%	<i>Hydrocarboniphaga effusa</i> (AY363244)	88%
B094	R	FR796636	Gammaproteobacteria	Uncultured bacterium clone D44 16S ribosomal RNA gene (EU234314)	upstream of Wang Yang River	98%	<i>Panacagrmonas perspica</i> (AB257720)	90%
B095	R	FR796637	Gammaproteobacteria	Uncultured sulfur-oxidizing symbiont bacterium partial 16S rRNA gene (AM935643)	hydrocarbon-contaminated soil	94%	<i>Halochromatium glycolicum</i> (X93472)	89%
B096	RD	FR796638	Gammaproteobacteria	Uncultured bacterium clone dr61 16S ribosomal RNA gene (AY540779)	gold mine south africa	97%	<i>Thiohalomonas denitrificans</i> (EF117913)	93%
B097	D	FR796639	Gammaproteobacteria	Uncultured gamma proteobacterium clone A19YC01RM small subunit (FJ568372)	soil early snow melt site B, Alpes	96%	<i>Steroidobacter denitrificans</i> (EF605262)	88%
B018 ^{HWW*}	D	FR796560	Nitrospira	Uncultured bacterium isolate SSCP band TW16-8R-16-6 16S ribosomal RNA, partial sequence (DQ077576)	drinking water distribution system HWW	100%	<i>Nitrospira moscoviensis</i> (X82558)	97%

B019	D	FR796561	Nitrospira	Uncultured bacterium DSSD16 16S ribosomal RNA gene, partial sequence (AY328715)	drinking water distribution system simulator	100%	Nitrospira moscoviensis (X82559)	99%
B027	D	FR796569	Planctomycetes	Uncultured planctomycete partial 16S rRNA gene, isolate OTU32/APA (AM902610)	subsurface thermal spring	97%	Zavarzinella formosa (AM162406)	88%
B028 ^a	D	FR796570	Planctomycetes	Uncultured bacterium clone F2_116X 16S ribosomal RNA gene, partial sequence (GQ262993)	simulated low level waste site	87%	Gemmata obscuriglobus (X56305)	90%
B029	D	FR796571	Planctomycetes	Uncultured bacterium clone FFCH1421 16S ribosomal RNA gene, partial sequence (EU135075)	soil from an undisturbed mixed grass prairie reserve	99%	Thermodesulfovibrio hydrogeniphilus (EF081294)	87%
B030	D	FR796572	Planctomycetes	Uncultured planctomycete clone PL09-10 16S ribosomal RNA gene, partial sequence (FJ844345)	water of high-mountain lake	97%	Pirellula staleyii (X81948)	87%
B031 ^a	D	FR796573	Planctomycetes	Uncultured bacterium NewOrleansYard3_YD3_032406_139 16S ribosomal RNA gene, partial sequence (FJ525164)	sediment	85%	Pirellula staleyii (X81948)	78%
B032	R	FR796574	Planctomycetes	Uncultured bacterium clone FW026-181 16S ribosomal RNA gene, partial sequence (EF692781)	sediment	91%	Subaequorebacter tamlense (AM293856)	85%
B087 ^a	R	FR796629	Planctomycetes	uncultured bacterium gene for 16S rRNA, partial sequence, clone:OS-54 (AB205985)	activated sludge	87%	Escherichia coli strain BEE25 16S ribosomal RNA gene (EF560792)	83%
B088 ^a	D	FR796630	Planctomycetes	Uncultured bacterium clone 168b1 16S ribosomal RNA gene (EF459840)	Baltic Sea sediment	83%	Nevskia soli (EF178286)	86%
B036 ^a	D	FR796578	TM6	Uncultured bacterium partial 16S rRNA gene, clone HA5-SRB-c056 (FM868200)	sediment	85%	Thiodictyon elegans (EF999973)	86%
B037	R	FR796579	TM6	Uncultured bacterium clone MD2902-B36 16S ribosomal RNA gene, partial sequence (EU385862)	subseafloor sediment of the South China sea	91%	Panacagrionas perspica (AB257720)	81%
B038	R	FR796580	TM6	Uncultured division TM6 bacterium clone NOS7.2WL 16S ribosomal RNA gene, partial sequence (AY043739)	forest cut-block surface organic matter	94%	Eubacterium rangiferina (EU124830)	82%
B039	RD	FR796581	TM6	Uncultured bacterium clone FGL7S_B80 16S ribosomal RNA gene, partial sequence (FJ437950)	Green Lake surface sediments	90%	Syntrophothermus lipocalidus (AB021305)	88%

B040	R	FR796582	TM6	Uncultured soil bacterium clone 331 16S ribosomal RNA gene, partial sequence (EU106159)	soil from radish rich area	91%	Rubrobacter radiotolerans (AJ243870)	88%
B041	R	FR796583	TM6	Uncultured bacterium partial 16S rRNA gene, clone a7-4 (FN594638)	biofilm from gold mine in Zloty Stok	93%	Desulfococcus multivorans (AF418173)	87%
B042	RD	FR796584	TM6	Uncultured candidate division TM6 bacterium partial 16S rRNA gene, clone CM1F08 (AM936568)	hydrocarbon-contaminated soil	92%	Eubacterium yurii subsp. schittka (AY533382)	88%
B043 ^a	R	FR796585	TM6	Uncultured bacterium clone I-GAC-3 16S ribosomal RNA gene, partial sequence (GQ452985)	water from drinking water treatment plant	88%	Hydrocarboniphaga effusa (AY363245)	90%
B044	D	FR796586	TM6	Uncultured bacterium clone PP4-50 16S ribosomal RNA gene, partial sequence (EU148985)	prepupa gut	94%	Wohlfahrtiimonas chitiniclastica (EU484335)	87%
B045	D	FR796587	TM6	Uncultured soil bacterium clone CWT SM03_G11 16S ribosomal RNA gene, partial sequence (DQ129127)	Coweeta forest soil	91%	Wohlfahrtiimonas chitiniclastica (EU484335)	86%
B046	R	FR796588	TM6	Uncultured candidate division TM6 bacterium clone DSR2W1u09 16S ribosomal RNA gene, partial sequence (EU635154)	showerhead swab	96%	Rubrobacter radiotolerans (AJ243870)	87%
B047	R	FR796589	TM6	Uncultured soil bacterium clone CWT SM03_G11 16S ribosomal RNA gene, partial sequence (DQ129127)	Coweeta forest soil	97%	Wohlfahrtiimonas chitiniclastica (EU484335)	86%

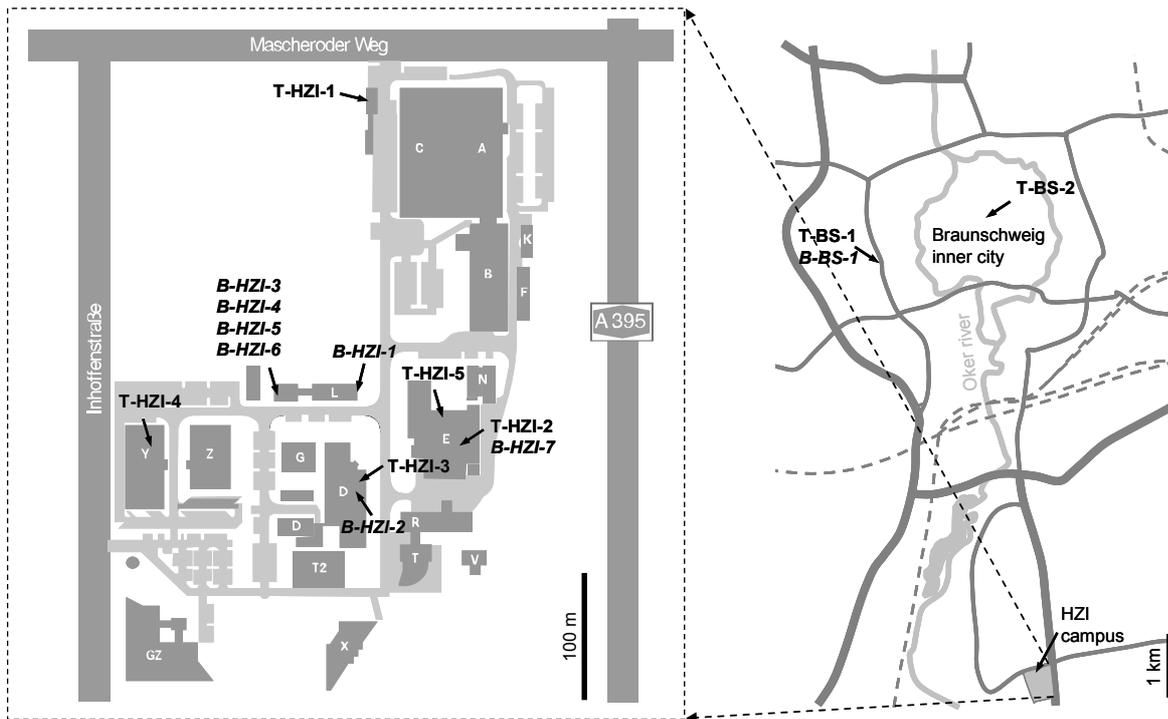
1 **Supplementary Table S5.** Number of phylotypes retrieved from bulk water and biofilms
 2 (presence/absence data). DNA: Number of phylotypes retrieved from DNA-based
 3 fingerprints. RNA: Number of phylotypes retrieved from RNA-based fingerprints.
 4 DNA+RNA: Number of phylotypes retrieved from both types of fingerprints.

5

Phylum / Class	Bulk water				Biofilm			
	DNA	RNA	DNA+RNA	Total	DNA	RNA	DNA+RNA	Total
<i>Acidobacteria</i>	0	0	0	0	4	5	0	9
<i>Actinobacteria</i>	7	0	0	7	6	0	0	6
<i>Bacteroidetes</i>	10	1	0	11	3	2	2	3
Candidate division TM6	0	3	0	3	5	9	2	12
<i>Chlamydiales</i>	0	0	0	0	3	8	1	10
<i>Chloroflexi</i>	0	0	0	0	4	1	0	5
<i>Cyanobacteria</i>	0	3	0	3	0	0	0	0
<i>Firmicutes</i>	0	0	0	0	3	2	0	5
<i>Nitrospira</i>	0	1	0	1	2	0	0	2
<i>Planctomycetes</i>	2	1	0	3	6	2	0	8
<i>Proteobacteria:</i>								
<i>Alphaproteobacteria</i>	2	3	0	5	16	16	4	28
<i>Betaproteobacteria</i>	5	4	0	9	6	6	2	10
<i>Gammaproteobacteria</i>	0	2	0	2	9	5	2	12
<i>Deltaproteobacteria</i>	0	0	0	0	1	1	0	2
Total	26	18	0	44	68	57	13	112

6

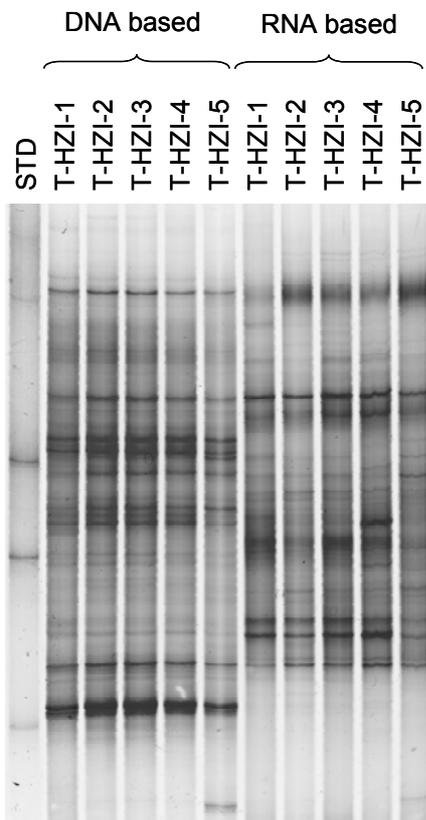
- 1 **Supplementary Figure S1.** Sampling locations in the Braunschweig area and at the campus
- 2 of the Helmholtz Centre of Infection research (HZI). Samples designated in bold italic are
- 3 biofilm samples (B-X), samples only in bold are bulk water samples (T-X).
- 4



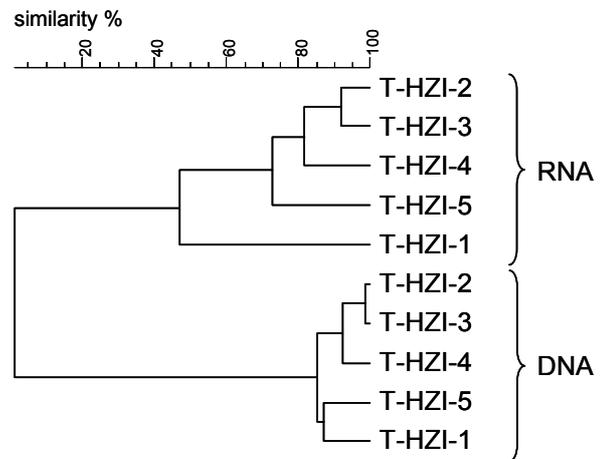
1 **Supplementary Figure S2.** (a) 16S rRNA gene based SSCP fingerprints of bulk water (left
 2 side, DNA based) and 16S rRNA-based SSCP fingerprints of bulk water (right side RNA
 3 based). (b) Comparative cluster analysis of DNA and RNA-based SSCP fingerprints of bulk
 4 water using Pearson algorithm.

5

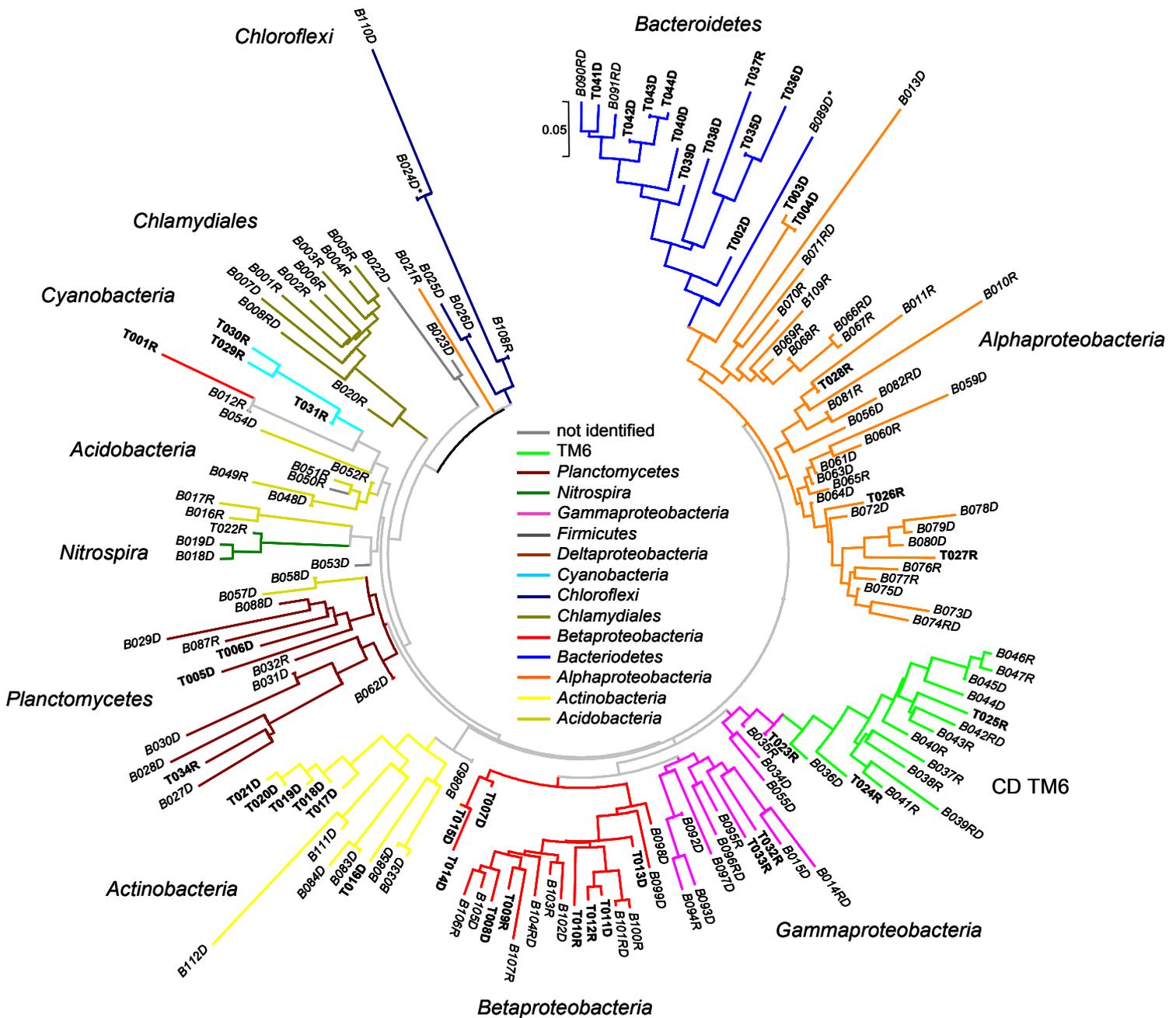
6 **a**



b

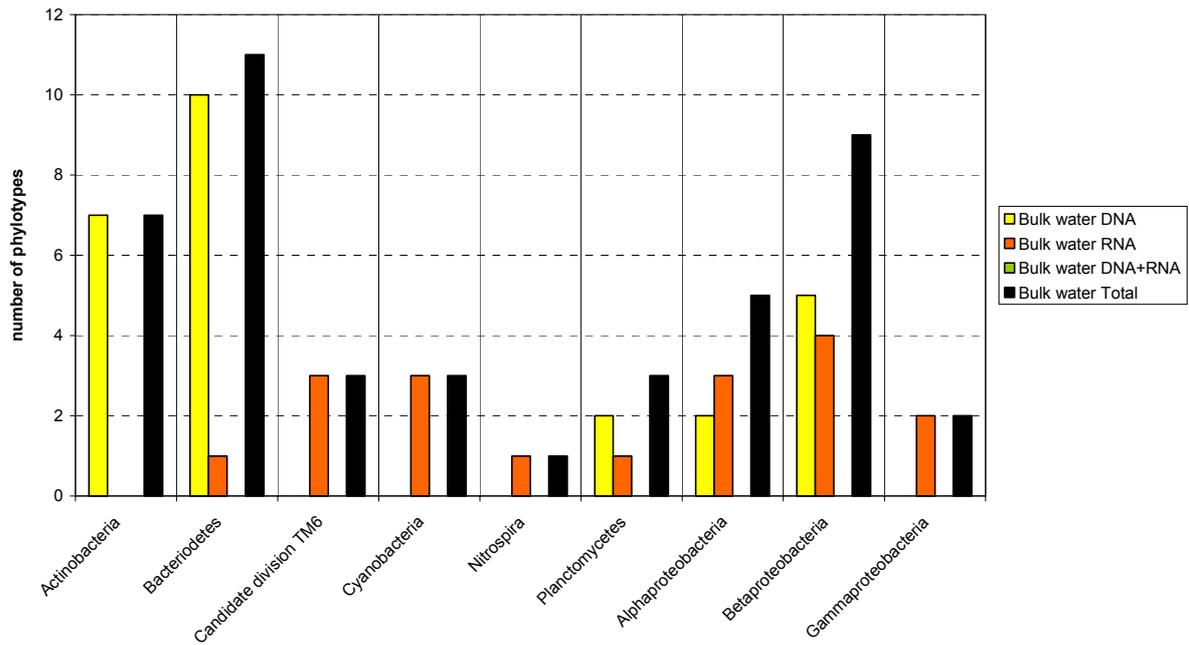


1 **Supplementary Figure S3.** Phylogenetic tree of all 16S rRNA gene sequences obtained from
 2 bulk water and biofilm phylotypes (detailed lists of the single phylotypes are given in
 3 Supplementary Table S3 and S4). Designation of sequences B: biofilm phylotype; T: bulk
 4 water phylotype; D originated from DNA-based fingerprint; R: originated from RNA-based
 5 fingerprint. All bulk water phylotypes are indicated in bold. The taxonomic tree was inferred
 6 using the Neighbor-Joining method. Evolutionary distances were computed using the
 7 Maximum Composite Likelihood method.
 8



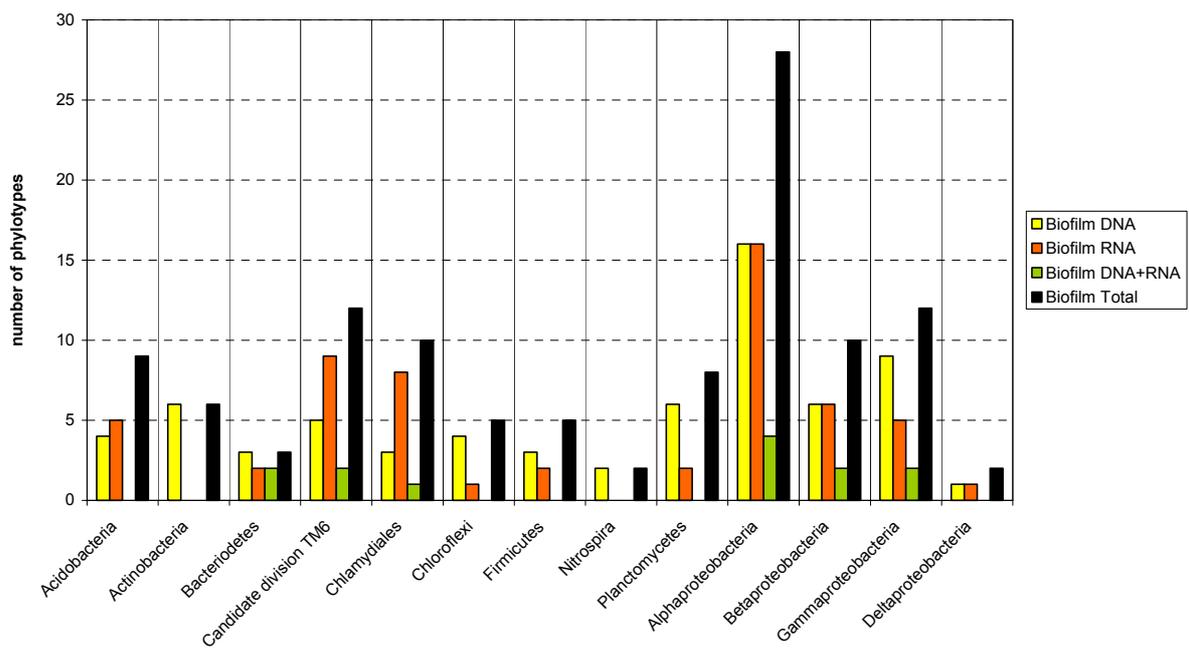
1 **Supplementary Figure S4.** a) Number of retrieved phylotypes from bulk water. b) Number
 2 of retrieved phylotypes from biofilm. DNA: Number of phylotypes found in DNA-based
 3 fingerprints. RNA: Number of phylotypes found in RNA-based fingerprints. DNA+RNA:
 4 Number of phylotypes found in both types of fingerprints.

5 a)



6

7 b)



8